CONTI

amplification from the pSVII-env plasmid (a gift from Dr. J. Sodroski, Dana-Farber Cancer Institute, Boston, MA). One fragment was generated by PCR with the synthetic oligonucleotide containing the SaII stite and the CCACC Kozak's sequence in front of the ATG codon (5'-AGAGTCGACCCACCATGAGAGTGAAGGAGA-3', sense) (SEQ ID NO:1), and the oligonucleotide (5' ACAGGTACCCCATAATAGACTGTGAC-3' antisense) (SEQ ID NO:2) containing the KpnI side, used for ligation with the second env fragment. The second fragment was derived by KpnI and BamHI digests of the pSVIII-env plasmid, and the third fragment was generated by PCR with the synthetic oligonucleotide containing the BamHI site at its 5' end (5'-AACGGATCCTTAGCACTTATCTGGG-3', sense) (SEQ ID NO:3) and the antisense primer (5'-TTGCGCGGCCGCTTATAGCAAAATCCTTTCC-3') (SEQ ID NO:4) containing the TAA stop codon followed by the *NotI* site. The three fragments were ligated into the SaII and NotI sites of the pSC11-based vector (a generous gift of Dr. L. Eisenlohr, Thomas Jefferson University, Philadelphia, PA) to generate plasmid pSC- Δ V3. A similar approach was used to generate plasmid with the WT env gene (pSC-WTP) using recombinant clone pIIIB (Hwang, et all, Science 253:71-74) kindly provided by Dr. B. Cullen (Howard Hughes Medical Institute, Duke University Medical Center, Durham, NC). Plasmids pSC- Δ V3 and pSC-WTP were used to generate vv- Δ V3 and vv-WTP by homologous recombination as described (Earl et al, 1990, J Virol. 64:2448-2451).--